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RESPONSES OF NONTARGET LEPIDOPTERA TO FORAY 48B® *BACILLUS THURINGIENSIS* VAR. *KURSTAKI* ON VANCOUVER ISLAND, BRITISH COLUMBIA, CANADA

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Abstract—Impacts of a gypsy moth (*Lymantria dispar*) eradication program on native, nontarget Lepidoptera were assessed in 1999, on southeastern Vancouver Island (BC, Canada). The microbial insecticide *Bacillus thuringiensis* var. *kurstaki* (*Btk*) was applied aerially over two areas totalling 12,805 ha on May 8, May 19, and June 8, 1999, at a dosage of 50 billion international units in 4.0 L/ha. Lepidoptera were collected from two host plant species: Garry oak (*Quercus garryana*) and common snowberry (*Symphoricarpos albus*). Lepidopteran larvae were collected from common snowberry foliage at 24 urban parks and from Garry oak foliage at 28 oak-dominated habitats, representing 12 and 14 replicates, respectively, of two treatments: unsprayed (reference) and sprayed (treatment). Prespray data were collected from March 25 to May 6 for *S. albus*, and from April 26 to May 6 for *Q. garryana*. Postspray data were collected from May 10 through June 15 for *S. albus* and from May 10 to July 6 for *Q. garryana*. The 15 most abundant lepidopteran species were analyzed statistically. However, the majority of species were collected infrequently, and, therefore were pooled for statistical analysis. After the *Btk* spray applications, 11 of the individual species and groups of uncommon species were found to be significantly less abundant in the treatment sites than in the reference sites. The effects of sample date were statistically significant on almost all groups of Lepidoptera analyzed, both before and after *Btk* spray applications, indicating temporal variation in lepidopteran abundance. Significant variation in diversity of members of the Lepidoptera, as a result of *Btk* spray application, was not detected on *S. albus* or *Q. garryana*. However, results showed significant variation in lepidoptera richness and abundance on both host plant species.

Keywords—Nontarget Lepidoptera Gypsy moth Garry oak *Bacillus thuringiensis kurstaki*

INTRODUCTION

The European gypsy moth (*Lymantria dispar* (L.)) has become a widespread and destructive forest and agriculture pest in North America since its introduction in the 1860s. Gypsy moth caterpillars are known to defoliate more than 300 species of plants [1], and the indirect effects, such as plant mortality, habitat destruction, and financial loss can be immense. The gypsy moth has been recorded at several sites throughout southwestern British Columbia, Canada, since the early 1970s, and in 1991 an Asian race of the moth was found. These populations have remained small and innocuous, likely because of eradication programs involving commercial formulations of the bacterium *Bacillus thuringiensis* var. *kurstaki* (*Btk*) [2].

Bacillus thuringiensis is a rod-shaped, aerobic bacterium that is found throughout most regions of the world [3,4] and has been isolated from insects, leaves, soil, and stored product dusts [5]. *Bacillus thuringiensis* var. *kurstaki* is toxic to only certain species of Lepidoptera that have a high midgut pH level that is capable of activating *Btk* toxins. Once ingested, these toxins lead to the perforation of the gut wall and cause death in susceptible species via gut paralysis or septicemia [6,7]. Although *Btk* formulations are less hazardous than the previously used broad-spectrum insecticides (i.e., carbaryl, acephate, and diflubenzuron), their environmental impacts should not be ignored. Concerns involve members of the Lepidoptera that are valued for their rarity or aesthetic beauty [8,9], their utility as biological control agents [10], or their importance as a food source for insectivores [11,12].

Several field studies assessing the direct impact of *Btk* sprays on nontarget Lepidoptera have concluded that *Btk* treatments applied during early spring can cause immediate reductions in the population density of some immature lepidopterans [13–18]. In spite of this common trend, data should be collected from numerous spray operations, because the effect of *Btk* depends on the formulation used, the number of applications, dosage, weather conditions, size of treatment areas, and other factors [19]. In addition, the responses of species of Lepidoptera may vary with the larval stage present during application of *Btk*, midgut protease and pH conditions, feeding behavior, and microhabitat preference [20]. Therefore, impact studies are a necessary component of *Btk* spray operations, because *Btk* exposure, which cannot be predicted, and lepidopteran community composition will determine the impact of the application. This paper reports on impacts of a gypsy moth eradication program that was conducted in 1999 on the southern tip of Vancouver Island (BC, Canada). The key objective was to monitor nontarget Lepidoptera on *Quercus garryana* Dougl. (Garry oak) and *Symphoricarpos albus* (L.) Blake (common snowberry) to determine the severity of population reductions resulting from three aerial applications of *Btk*.

MATERIALS AND METHODS

Study area

Immature lepidopterans were monitored on Garry oak and common snowberry (snowberry) throughout much of the Vic-

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Table 1. Dates of sampling cycles and *Bacillus thuringiensis kurstaki* (*Btk*) spray applications during the 1999 gypsy moth eradication program on southern Vancouver Island (BC, Canada)

	Dates		
	Sampling for snowberry	Sampling for Garry oak	*Btk* spray applications
Prespray period	March 25–April 1	April 26–30	
	April 1–6	May 2–6	
	April 8–15		
	April 16–22		
	April 26–30		
	April 30–May 6		
Postspray period	May 10–13	May 10–13	May 8
	May 20–23	May 21–24	May 19
	May 31–June 3	June 1–4	June 8
	June 12–15	June 16–19	
		July 2–6	

toria metropolitan area on southern Vancouver Island. The population of Greater Victoria is about 325,000, and approximately 58% of the land area is green space (11% parks and protected areas, 25% unprotected area, and 12% agricultural land reserve). Victoria lies in the rain shadow of the Olympic Mountain Range; mean annual precipitation ranges from 647 to 1,263 mm, and mean annual temperature ranges from 9.2 to 10.5°C (Victoria International Airport, Sydney BC, Canada).

Btk *application*

Bacillus thuringiensis var. *kurstaki* (Foray 48B®, Valent Biosciences, Libertyville, IL, USA) was applied to two spray zones in the Greater Victoria area by fixed-winged aircraft, at a rate of 50 billion international units in 4.0 L/ha. Both zones were sprayed on May 8, May 19, and June 8, 1999, between 0530 and 0700 h. Spray droplets on Kromekote® cards (Smart papers LLC, Hamilton, OH, USA; two to four cards per site) confirmed spray penetration to the ground at all treatment sites. No deposits were detected at study sites located outside the defined spray-zone boundaries.

Study sites and sampling procedures

Snowberry and Garry oak were selected because both species are abundant and host large numbers of immature lepidopterans. Snowberry has an earlier phenology than Garry oak, and was, therefore, used to study nontarget lepidopteran species that feed during the early spring before Garry oak leaves are fully expanded. Snowberry is a small- to medium-sized, erect, deciduous shrub that is widespread throughout Greater Victoria. Garry oak is the only oak native to British Columbia, where it occurs in two general habitat types: open meadow with large healthy oaks, and rocky knoll with typically gnarled and stunted oaks [21].

A total of 24 sites (each ~ 1–3 ha) were chosen to sample snowberry. Twelve sites were located within the larger spray zone in Greater Victoria and paired with 12 unsprayed reference sites that were similar in elevation, canopy cover, and proximity to the ocean. Within each snowberry site, 10 contiguous patches (each ~ 100–200 m^2) of snowberry plants were selected, and at each sampling date, one plot was randomly selected, without replacement, and lepidopterans were sampled from the snowberry plants. Prespray sampling began as soon as the majority of leaves started to flush (Table 1). Six prespray and four postspray samples were collected with a novel method, by shaking 50 snowberry branches, one by one, above a white tray (43.2 × 50.8 × 7.8 cm [17 × 20 × 3 in.]) and by visually searching the tray for caterpillars.

Garry oak study sites were selected in seminatural parks and private estates according to the criteria reported by Sopuck et al. [12]. Twenty-eight Garry oak sites were sampled: 13 were located within the larger (12,203-ha) spray zone in Greater Victoria, and 1 site was within the smaller (602-ha) separate spray zone in Brentwood Bay (BC, Canada). The sampling method was similar to that of Miller [13]. Prespray sampling began as soon as the majority of leaf buds on Garry oaks started to flush (Table 1). Each sampling effort required 4 d to complete, and paired treatment and reference sites were sampled on the same day. At each site, three randomly selected trees were sampled at each sampling date by vigorously shaking one branch for 10 s with a 2-m wooden pole. Branches were selected only if they were not sampled previously, they were within 2 m of the ground, their base diameter was greater than 4 cm, and they possessed at least 20 leaf clusters. Leaf clusters were counted on all branches to ensure that equal amounts of foliage were sampled in the treatment and reference plots. The dislodged caterpillars fell onto a 2 × 3-m white nylon sheet, from which they were removed and immediately placed in 70% ethanol. Larvae that could not be identified in the larval stage were sorted into morphotypes, each of which was presumed to be a distinct species.

Statistical analysis

Lepidopteran abundance data were expressed as number per 10 branches for common snowberry, and number per three branches for Garry oak, and were logarithmically transformed ($\log(x + 1)$) to improve normality. Two-factor analysis of variance (ANOVA) models were used to examine the variation among treatments and sample dates in lepidopteran total abundance and species richness and the abundance of selected taxa and lepidopteran groups. The final sampling cycle for Garry oak could not be included in the postspray ANOVA because the branches used in this seventh cycle were selected after the original randomization of branches involving the first six cycles. Therefore, an independent sample *t* test was used to analyze data from the seventh sample.

Because only two *Btk* spray zones were included, this study could not be well replicated. To minimize the effects of pseudoreplication [22], each Garry oak and snowberry treatment site was paired with a similar unsprayed reference site located outside the aerial spray boundaries. The criteria for pairing included elevation, aspect, proximity to the ocean, and qualities of the shrub layer as described by Sopuck et al. [12]. Because each site within the spray zone was matched with a reference site that was judged to have similar landscape characteristics, the sample design approximated replication because the spray plots were large and the pesticide effects greatly exceeded potentially confounding variables such as weather. In the future, data from this study may be combined with data from other pseudoreplicated studies in a meta-analysis, resulting in a more powerful test of the null hypothesis (H_0) [23].

RESULTS

Snowberry

Throughout the study, 10,050 lepidopteran larvae were collected from 12,000 snowberry branches (Table 2). Only 2 of

Table 2. Comparison of species richness and abundance of immature lepidopterans on common snowberry and Garry oak between the treatment and control sites in Greater Victoria (BC, Canada), April 26 through June 19, 1999

	Total abundance of larvae		Species or morphotype richness	
	Treatment sites	Control sites	Treatment sites	Control sites
Prespray snowberry	3,845	3,813	25	21
Postspray snowberry	993	1,399	20	25
Prespray Garry oak	2,056	2,830	26	24
Postspray Garry oak	3,551	7,781	46	52

36 morphotypes were identified at the species level (Table 3). The most abundant lepidopteran species hosted by snowberry was the snowberry moth (*Euceratia castella* (Walsingham); Plutellidae), which accounted for 47.7% of all caterpillars collected. The snowberry moth reached its peak abundance by April 16 to 22 and declined steadily, disappearing by June 8 (Fig. 1). The winter moth (*Operophtera brumata* (Linnaeus); Geometridae), which was the second most abundant species, accounted for approximately 35.9% of total abundance. The winter moth did not reach an obvious peak in abundance on snowberry. The remaining morphotypes were not abundant enough for statistical analysis, so they were divided into two groups: uncommon Lepidoptera (morphotypes that were collected 100–500 times throughout the study) and very uncommon Lepidoptera (morphotypes that were collected fewer than 100 times throughout the study). The uncommon and very uncommon lepidopteran groups accounted for approximately 10.4% and 6.0% of total abundance, respectively, and both groups had multiple peaks in abundance. Total lepidopteran abundance at the treatment and reference sites increased steadily from March 25 through April 22 and decreased steadily from April 30 through June 15. The first *Btk* spray application occurred two to three weeks after peak abundance of the guild of immature lepidopterans on snowberry.

During the prespray period, 3,845 larvae and 25 morphotypes were found in the treatment sites and 3,813 larvae and 21 morphotypes were found in the reference sites. Analysis of variance models showed that lepidopteran species richness; total abundance; and densities of the winter moth, uncommon Lepidoptera, and very uncommon Lepidoptera were not significantly different between the treatment and reference sites. However, mean abundance of the snowberry moth was significantly higher in the treatment sites than in the reference sites before *Btk* applications ($p \leq 0.01$, $F = 9.40$). The snowberry moth also was significantly affected by time and a time–treatment interaction ($p \leq 0.01$, $F = 8.57$), suggesting that its phenology may have varied between the treatment and reference plots (Table 4).

During the postspray period, 993 larvae and 20 morphotypes were found on snowberry in the treatment sites and 1,399 larvae and 25 morphotypes were found in the reference sites. The abundance of the snowberry moth was not significantly different between the treatment and reference sites. However, species richness ($p \leq 0.01$, $F = 33.88$); total abundance ($p \leq 0.01$, $F = 24.33$); and the densities of the winter moth ($p \leq 0.01$, $F = 13.68$), uncommon Lepidoptera ($p \leq 0.01$, $F = 18.04$), and very uncommon Lepidoptera ($p \leq 0.01$, $F = 10.49$) were significantly lower in the *Btk*-sprayed sites than in the reference sites. In the postspray ANOVA for the snowberry moth and total abundance, significant one-way interactions were found between treatments and sample cycles, probably because a lag time occurred between the *Btk* application and the mortality of affected larvae.

Garry oak

Throughout the study, 16,218 lepidopteran larvae were collected from 588 Garry oak branches. Species-level identifications were made on 99.4% of the specimens; however, species names could only be placed on 33 of 57 morphotypes. The most abundant lepidopteran species on Garry oak was the winter moth, which accounted for 40.3% of total abundance. The winter moth reached a maximum abundance in the samples from May 10 through 13 (Fig. 2). *Chionodes trichostola* (Gelechiidae), which was the second most abundant species, accounted for 13.2% of total abundance. *Chionodes trichostola* increased its population steadily throughout the sampling period, peaking in abundance from July 2 through 6. Twelve other species were relatively abundant, and together comprised 44.4% of total abundance. The remaining species were collected in too few numbers for statistical analysis, so they were analyzed as a group (uncommon species), which accounted for approximately 2.1% of total abundance. The date of peak abundance varied among the different species; only five species reached their peak abundance before the first *Btk* spray application, compared with 33 during the postspray period. Total lepidopteran abundance at treatment and reference sites increased steadily during the early spring, and peaked at May 21 through 24. Total lepidopteran abundance declined sharply in the treatment sites between May 24 and June 1, but declined only slightly in the reference sites.

During the prespray period, 2,056 larvae and 26 morphotypes were collected in the treatment sites compared with 2,830 larvae and 24 morphotypes in the reference sites. *Hydriomena nubilofasciata* ($p \leq 0.01$, $F = 52.46$) and *Erranis tiliaria vancouverensis* ($p \leq 0.01$, $F = 7.24$) were significantly more abundant in the reference sites than in the treatment sites during the prespray period. The remaining common species, the uncommon species group, and total lepidopteran abundance were not found to differ between the treatment and reference sites during the prespray period. The interaction between treatment and sample cycle was statistically significant only for *Spilonota ocellana* ($p \leq 0.05$, $F = 4.34$) during the prespray period, and not for any other species.

During the postspray period, 3,551 larvae and 46 morphotypes were collected from Garry oak in the treatment sites compared with 7,781 larvae and 52 morphotypes in the reference sites. Densities of most of the abundant species (9 of 14) and the uncommon species group ($p \leq 0.05$, $F = 5.75$), total lepidopteran abundance ($p \leq 0.01$, $F = 36.49$), and species richness ($p \leq 0.01$, $F = 26.41$) were found to be significantly lower in the treatment sites than the reference sites during the postspray period. Total lepidopteran abundance remained significantly lower in the treatment sites than the reference sites until at least July 2 through 6. In the postspray period, significant interactions occurred in *S. ocellana* ($p \leq 0.05$, $F = 1.63$), *Telphusa sedulitella* ($p \leq 0.05$, $F = 4.94$), *Ypsolopha cervella* ($p \leq 0.05$, $F = 3.01$), *Pandemis ceresana* ($p \leq 0.05$, $F = 3.22$), *Ditula angustiorana* ($p \leq 0.05$, $F = 2.17$), *Orthosia pacifica* ($p \leq 0.05$, $F = 0.64$), and total abundance. The significant interactions during the postspray period may be explained by a lag time between the spraying of *Btk* and the death of affected larvae.

Table 3. Immature lepidopteran species composition on Garry oak in Greater Victoria (BC, Canada), April 26 through July 6, 1999

Host plant	Taxa	Total abundance	Percent abundance
Snowberry	Plutellidae		
	Euceratia castella	5,136	47.73
	Geometridae		
	Operophtera brumata (winter moth)	3,863	35.90
	Uncommon Leipdoptera	1,116	10.37
	Very uncommon Lepidoptera	646	6.00
Garry oak	Arctiidae		
	Cleminsia albata	1	0.01
	Gelechiidae		
	Chionodes trichostola	2,135	13.16
	Telphusa sedulitella	1,118	6.89
	Geometridae		
	Operophtera brumata[a]	6,540	40.33
	Hydriomena nubilofasciata[a]	1,623	10.01
	Erranis tiliaria vancouverensis	174	1.07
	Venusia pearsalli	46	0.28
	Coniodes plumogeraria	11	0.07
	Cyclophora dataria	7	0.04
	Biston cognataria[b]	5	0.03
	Eupithecia spp.	4	0.02
	Lambdina fiscellaria somniaria[a]	4	0.02
	Pero sp.[b]	2	0.01
	Hemithea aestivarea[c]	1	0.01
	Neoalcis californiaria	2	0.01
	Unknowns (16 morphotypes)	65	0.40
	Gracillaridae		
	Caloptilia sanguinella	929	5.73
	Hesperidae		
	Erynnis propertius[b]	1	0.01
	Lasiocampidae		
	Phyllodesma americana	3	0.02
	Lymantriidae		
	Orgia antiqua badia	4	0.02
	Noctuidae		
	Orthosia pacifica	82	0.51
	Catocala aholibah	60	0.37
	Egira simplex	49	0.30
	Aseptis binotata	3	0.02
	Unknowns (six morphotypes)	14	0.09
	Oecophoridae		
	Carcina quercana	21	0.13
	Olethreutidae		
	Spilonota ocellana	1,691	10.43
	Epinotia emarginana	22	0.14
	Plutellidae		
	Ypsolopha cervella	337	2.08
	Torticidae		
	Choristoneura rosaceana[a]	268	1.65
	Archips rosana[a]	456	2.81
	Pandemis cerasana[a]	302	1.86
	Ditula angustiorana[a]	155	0.96
	Decodes fragarianus	60	0.37
	Argyrotaenia citrana	4	0.02
	Unknowns (one morphotype)	1	0.01
	Unidentified family	18	0.11

[a] Introduced species and pests.
[b] Rare potentially vulnerable species.
[c] New record for Garry oak.

DISCUSSION

In general, a significant difference was lacking between Lepidoptera in the treatment and reference sites during the prespray period in terms of the abundance and species richness of larvae on snowberry and Garry oak. The only exceptions were *H. nubilofasciata* and *E. tiliaria vancouverensis*, which were more abundant on oak in the reference sites during the prespray period, and the snowberry moth, which was more abundant on snowberry in the treatment sites during the prespray period. However, after the *Btk* was applied, the uncommon species groups on snowberry and Garry oak were significantly more abundant in the reference sites than in the treatment sites, as were most of the common species. Lepidopteran species richness also dropped in sprayed sites relative to reference sites on both host plant species. Therefore, the conclusion is made that the 1999 gypsy moth eradication pro-

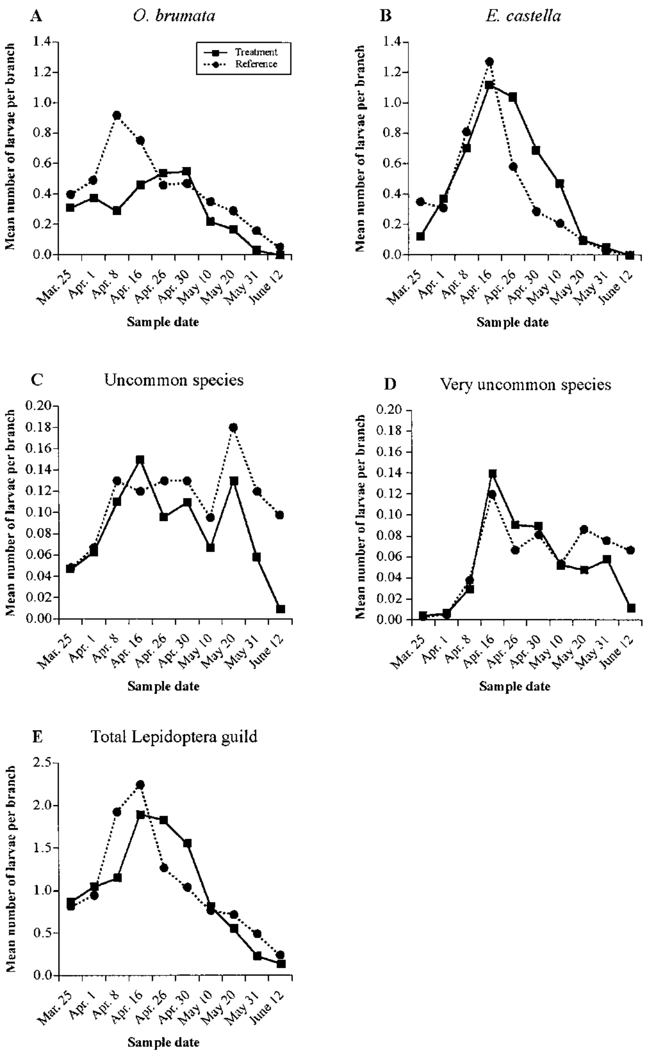

Fig. 1. Abundance of caterpillars (average number per branch) collected from snowberry in the treatment and control plots: (**A**) winter moth (*Operophtera brumata*), (**B**) snowberry moth (*Euceratia castella*), (**C**) uncommon species (morphotypes collected 100–500 times), (**D**) very uncommon species (morphotypes collected fewer than 100 times), and (**E**) total abundance of all species collected. The microbial insecticide *Bacillus thuringiensis* var. *kurstaki* was applied on May 8, May 19, and June 8, 1999.

gram on southern Vancouver Island was generally detrimental to nontarget members of the order Lepidoptera that were present in the feeding stage on snowberry and Garry oak when the *Btk* was applied. Moreover, because numbers of larvae were reduced on snowberry, a shrub with an early phenology, the majority of lepidopterans in the spray zones apparently were unable to complete their larval stage before the *Btk* spraying began. However, five of the species affected were introduced from Europe, and three of them (*O. brumata*, *H. nubilofasciata*, and *P. cerasana*) can be considered pests on Garry oak. Thus, reductions of these unwanted lepidopterans may be considered a secondary benefit of the *Btk* spraying.

Life-history characteristics appeared to protect three of the unaffected species. *Spilonota ocellana* was not significantly reduced in the treated sites, probably because it has an early phenology that caused it to reach the pupal stage before obtaining a lethal dose of *Btk*. *Chionodes trichostola* remained common in treated sites, probably because of an extended period of larval emergence. By the time *Btk* was first applied, the abundance of the snowberry moth was 64% lower than peak abundance, a result of completed larval development and/or natural mortality. Thus, a type II error, nonrejection of H_0 when the alternative hypothesis (H_1) is true, may have been committed because the chance of detecting a treatment effect would have declined as the abundance of immature lepidopterans declined. On the other hand, the reason for nonrejection of H_0 may be that the snowberry moth avoided *Btk* residues by constructing, and feeding within, leaf-rolls. Several authors have observed low levels of mortality among microlepidopterans after pesticide treatments and have attributed this phenomenon to their tendency to feed in concealed microhabitats where they are unlikely to encounter lethal dosages [15,17,18]. *Ditula angustiorana* was not significantly lower in abundance in the *Btk*-sprayed sites than in the reference sites but this result could reflect its relatively low abundance, and hence the relatively low power of its ANOVA. *Choristoneura rosaceana* and *Archips rosana* are the only other abundant species that were not found to be significantly reduced in number during the postspray period. When combined together, *A. rosana* and *C. rosaceana* were significantly less abundant in the treatment sites than in the reference sites, indicating that the *Btk* probably impacted them.

The impact of *Btk* on lepidopteran total abundance appeared more severe after the third *Btk* spray application than after the first two applications, probably because the rate of natural population growth (i.e., the emergence of new larvae) exceeded the rate of death from *Btk* during the early portion of the postspray period. In addition, the higher efficacy of *Btk* late in the season may reflect delayed responses of insects, cumulative effects of multiple sprays, and seasonal changes in oak foliage. For example, the dose of *Btk* on leaves can be diluted during the early spring by the expansion of growing leaves [24] and tannins, which increase with leaf age, can interact with *Btk* to make it more toxic in some instances [25].

Although some of the ANOVA models indicated significant interactions between the main effects, the *F* statistics for the main effects can be valid because the experimental design was orthogonal. The cause of the significant interactions appears to be a delayed response of some species to the *Btk* spray applications, possibly confounded with a slightly later phenology of some lepidopterans in the treatment sites relative to the reference sites. The significant interaction in the prespray ANOVA model of *S. ocellana* is puzzling, although type I errors are not uncommon when many ANOVA models are run. The interactions could also be influenced by *Btk* concentrations changing over time [20] and larval sensitivity to *Btk* changing with larval maturity [8].

Most of the lepidopteran species were collected infrequently and more than one half of the known lepidopteran fauna on snowberry and Garry oak was absent in the study samples, probably because of naturally low population densities. Thus, the suggestion is made that many of the native lepidopteran species on these two host plants are uncommon on southern Vancouver Island. The decline of the uncommon species groups in the *Btk* spray zones suggests that some of the rare species in this study may have been severely impacted by the *Btk* applications. Rare species are of special concern because they may be more easily extirpated by disturbances such as *Btk* spraying than common species [14]. Nevertheless, conservation considerations also should include reductions of common species because they can contribute the most to certain ecosystem processes [26]. The significant decline of the

Table 4. Results of two-factor analysis of variance models for effects of treatment (*Bacillus thuringiensis kurstaki* spray vs no spray) and sample date on species richness, total abundance, and selected taxa

	Treatment		Sampling date		Treatment–sampling date interaction	
Dependent variable	*F*	*df*	*F*	*df*	*F*	*df*
Prespray snowberry						
Euceratia castella	9.40**	1	8.57**	5	2.50*	132
Operophtera brumata	2.33	1	1.36	5	1.28	132
Uncommon	0.24	1	2.90*	5	0.54	132
Very uncommon	1.18	1	21.00**	5	0.59	132
Total abundance	0.02	1	9.68**	5	1.97	132
Species richness	0.00	1	10.29**	5	0.61	132
Prespray Garry oak						
Operophtera brumata	1.66	1	0.00	1	0.00	52
Chionodes trichostola	1.64	1	0.82	1	0.57	52
Spilonota ocellana	0.53	1	1.97	1	4.34*	52
Hydriomena nubilofasciata	11.90**	1	0.54	1	0.66	52
Telphusa sedulitella	0.17	1	8.83	1	1.09	52
Caloptilia sanguinella	1.34	1	0.01	1	0.00	52
Archips rosana	2.78	1	0.43	1	0.57	52
Ypsolopha cervella	0.11	1	0.70	1	0.46	52
Pandemis cerasana	0.54	1	1.71	1	3.53	52
Choristoneura rosaceana	2.68	1	0.43	1	0.55	52
Erranis tiliaria vancouverensis	9.17**	1	0.26	1	0.26	52
Ditula angustiorana	0.16	1	0.79	1	3.58	52
Decodes fragarianus	1.00	1	1.00	1	1.00	52
Orthosia pacifica	0.10	1	3.74	1	0.10	52
Uncommon species (pooled)	0.22	1	1.51	1	0.30	52
Total abundance	2.70	1	0.66	1	0.53	52
Species richness	0.08	1	2.39	1	0.81	52
Postspray snowberry						
Euceratia castella	2.05	1	58.56**	3	3.10*	88
Operophtera brumata	13.68**	1	20.00**	3	0.63	88
Uncommon	18.04**	1	9.92**	3	1.64	88
Very uncommon	10.49**	1	3.00*	3	2.04	88
Total	24.33**	1	57.22**	3	4.78**	88
Species richness	33.88**	1	17.17**	3	2.69	88
Postspray Garry oak						
Operophtera brumata	10.52**	1	52.24**	3	1.61	104
Chionodes trichostola	2.18	1	1.39	3	3.88	104
Spilonota ocellana	0.18	1	41.88**	3	1.63*	104
Hydriomena nubilofasciata	52.46**	1	24.94**	3	1.87	104
Telphusa sedulitella	9.81**	1	6.92**	3	4.94*	104
Caloptilia sanguinella	5.21*	1	37.94**	3	1.14	104
Archips rosana	1.99	1	6.57**	3	1.42	104
Ypsolopha cervella	24.55**	1	8.32**	3	3.01*	104
Pandemis cerasana	17.07**	1	2.78*	3	3.22*	104
Choristoneura rosaceana	2.47	1	6.62**	3	1.17	104
Erranis tiliaria vancouverensis	7.24**	1	6.52**	3	1.80	104
Ditula angustiorana	0.33	1	4.33**	3	2.17*	104
Decodes fragarianus	5.61*	1	0.78	3	0.15	104
Orthosia pacifica	12.49**	1	6.26**	3	0.64*	104
Uncommon species (pooled)	5.75*	1	7.17**	3	0.51	104
Total abundance	36.49**	1	33.74**	3	3.12*	104
Species richness	26.41**	1	22.19**	3	2.08	104

* $p \leq 0.05$, ** $p \leq 0.01$.

winter moth and the apparent lack of *Btk* impact on the snowberry moth are particularly interesting because these lepidopterans typically occur in large numbers on Garry oak and snowberry, respectively, which are dominant plant species in many habitats on southern Vancouver Island.

The total number of caterpillars in the spray-zones was reduced by an estimated 29% and 53% on snowberry and Garry oak, respectively. These reductions may have been harmful to other animals that depend on immature lepidopterans as a food source during the spring. However, a concurrent study of birds [12] using 21 of the Garry oak sites showed no impact of *Btk* spraying on the relative abundance of most (41 of 42) adult birds. The spotted towhee (*Pipilo maculates*) was the only potentially harmed bird, but in 2000 it was equally abundant in treated and reference plots. This demonstrates that the degree of larval mortality is less important to predators than is the absolute abundance of remaining individuals. Whether or not other types of predators (i.e., small mammals) were harmed is unknown.

The apparent reduction of lepidopteran species richness on Garry oak in the treatment sites after the *Btk* spray applications should be interpreted carefully because most species were col-

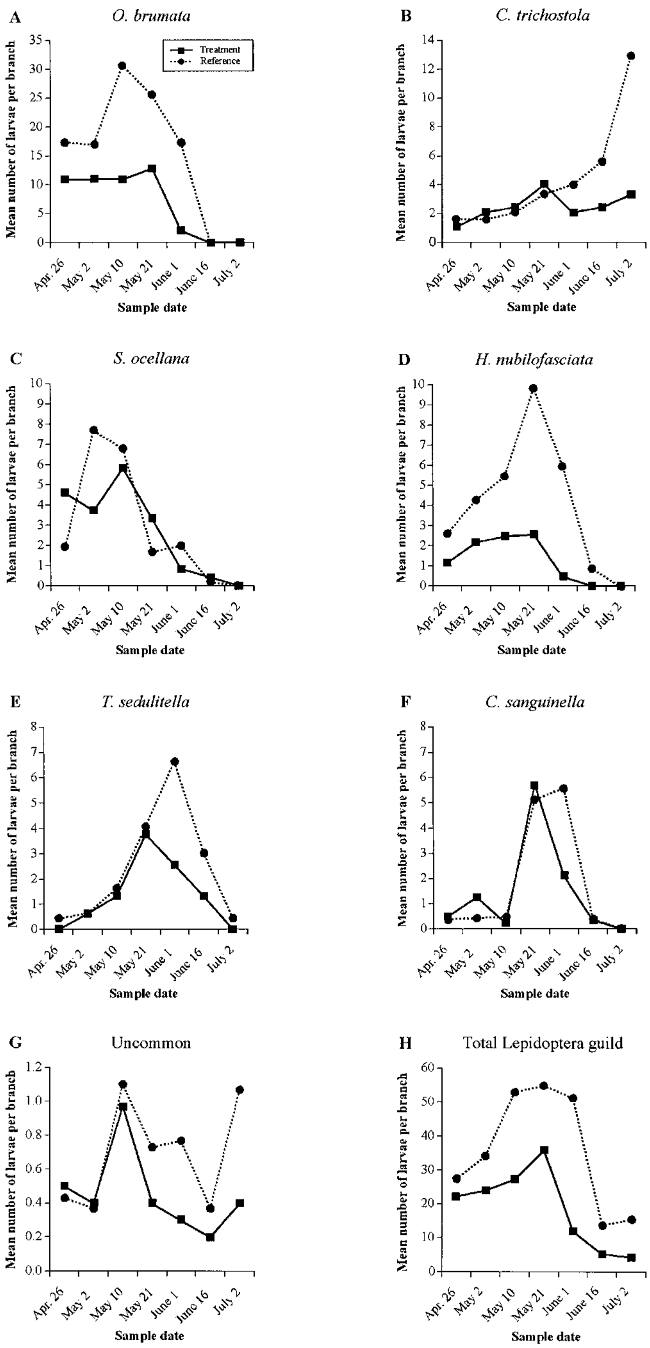

Fig. 2. Abundance of caterpillars (average number per branch) collected from Garry oak in the treatment and control plots: (**A**) through (**F**) the six most abundant species (*Operophtera brumata*, *Chionodes trichostola*, *Spilonota ocellana*, *Hydriomena nubilofasciata*, *Telphusa sedulitella*, and *Caloptilia sanguinella*), (**G**) the least abundant species (as a group), and (**H**) the total abundance of all species collected. The microbial insecticide *Bacillus thuringiensis* var. *kurstaki* was applied on May 8, May 19, and June 8, 1999.

lected infrequently. As a result, the reduction of species in the treatment site samples probably reflects diminished abundance rather than extirpation. Species richness is a valuable parameter when all the nontarget species of concern are collected frequently and can easily be identified, but the sampling effort necessary to achieve this may be impractical and possibly even harmful to local populations. Several authors [13,15,16] have reported that *Btk* causes immediate reductions in lepidopteran species richness, but their results may reflect diminished abundance rather than extirpation of uncommon species. Thus, species richness data must be interpreted carefully because they are influenced by sample size and species composition [27]. In addition, species richness indices can be insensitive to local extinctions when they are analyzed statistically, and in some cases local extinctions may not be apparent in short-term studies [28]. For example, if a lepidopteran species is depleted to the point that adults have difficulty locating mates, a lag time could occur between the initial disturbance and the point of extirpation [26].

The reductions of nontarget lepidopterans observed in this study may impose further stress on Garry oak ecosystems on southern Vancouver Island. Natural Garry oak habitat has been almost completely destroyed in British Columbia because of urbanization, industrialization, agriculture, and the invasions of exotic species [21]. The remaining fragments of the original habitat contain about 75% of British Columbia's endangered, threatened, and vulnerable plant taxa, and they are considered to be at high risk with respect to biodiversity conservation [21]. One butterfly species (*Erynnis propertius*) and three subspecies (*Coenonympha californica insulana*, *Euchloe ausonides isulanus*, and *Euphydryas editha taylori*) are found only in oak meadows and rocky knolls. Only *E. propertius* was present in the study samples. *Coenonympha californica insulana* occurs on Vancouver Island but is red listed by the province of British Columbia; *E. ausonides isulanus* is listed by the Committee on the Status of Endangered Wildlife in Canada as extirpated from Vancouver Island with only a single known population on San Juan Island; and *E. editha taylori* is listed by the Committee on the Status of Endangered Wildlife in Canada as endangered with no extant populations on Vancouver Island. Moths also could be at risk of becoming locally extinct, but a paucity of information exists on this large and diverse taxon.

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